

AMENDMENTS TO THE CLAIMS

Claim 1 (Currently amended): A method to stably modulate the expression of a target gene in a plant ~~cell tissue, seed, or whole plant~~, which method comprises the steps of:

a) introducing into the~~providing~~ plant cells with a synthetic zinc finger protein that is stably expressed, said zinc finger protein being capable of specifically binding to a target nucleotide sequence, or a complementary strand thereof, within a target gene, wherein said target nucleotide sequence comprises 18 nucleotides and wherein said zinc finger protein is a hexadactyl zinc finger protein, wherein said zinc finger protein comprises one individual zinc finger protein with mutations at one or more of the base-contacting positions; and

b) allowing said zinc finger protein binding to bind to said target nucleotide sequence, whereby the expression of said target gene in said plant ~~tissue, seed, or whole plant cell~~ is stably modulated.

Claim 2 (Canceled)

Claim 3 (Currently amended): The method of claim 1, wherein a nucleotide sequence encoding the zinc finger protein is expressed in the plant ~~tissue, seed, or whole plant cell~~ and the plant ~~tissue, seed, or whole plant cell~~ are maintained under conditions such that the expressed zinc finger protein binds to the target nucleotide sequence and regulates the expression of the target gene in the plant ~~tissue, seed, or whole plant cell~~.

Claim 4 (Currently amended): A method to stably modulate the expression of a target gene in ~~[[a]] plant cell~~cell, which method comprises the steps of:

a) providing introducing into the plant cells cell with an expression system for a synthetic zinc finger protein, said zinc finger protein being capable of specifically binding to a target nucleotide sequence, or a complementary strand thereof, within a target gene, wherein said target nucleotide sequence comprises 18 nucleotides and wherein said zinc finger protein is a hexadactyl zinc finger protein, wherein said zinc finger protein comprises one individual zinc finger DNA binding site with mutations at one or more of the base-contacting positions; and

b) culturing said plant cellcell under conditions wherein said zinc finger protein is stably produced and binds to said target nucleotide sequence,

whereby expression of said target gene in said plant ~~eells~~ cell is stably modulated.

Claim 5 (Previously presented): The method of claim 1, wherein the target nucleotide sequence is endogenous or exogenous to the target gene.

Claim 6 (Previously presented): The method of claim 1, wherein the target nucleotide sequence is upstream of, downstream of, or within the coding region of the target gene.

Claim 7 (Previously presented): The method of claim 1, wherein the target nucleotide sequence is DNA, RNA, PNA or a combination thereof.

Claim 8 (Previously presented): The method of claim 1, wherein the target nucleotide sequence is a promoter of a regulatory protein.

Claims 9-10 (Canceled)

Claim 11 (Previously presented): The method of claim 1, wherein the targeted nucleotide sequence is of the formula (GNN)₆, and wherein N is any one of the A, T, C or G.

Claim 12 (Canceled)

Claim 13 (Previously presented): The method of claim 1, wherein the target nucleotide sequence is endogenous to the plant but is in a non-naturally-occurring location.

Claim 14 (Currently amended): The method of claim 1, wherein the plant ~~eells~~ cell ~~comprise~~ comprises at least two copies of the same or different target nucleotide sequence(s).

Claim 15 (Previously presented): The method of claim 14, wherein each target nucleotide sequence is located within a different target gene, whereby more than one different target genes are modulated.

Claim 16 (Previously presented): The method of claim 1, wherein the target gene encodes a product that affects biosynthesis, modification, cellular trafficking, metabolism and degradation of a peptide, a protein, an oligonucleotide, a nucleic acid, a vitamin, an oligosaccharide, a carbohydrate, a lipid, or a small molecule.

Claim 17 (Previously presented): The method of claim 16, wherein the target gene encodes a protein or an RNA.

Claim 18 (Previously presented): The method of claim 1, wherein the target gene encodes myoinositol 1-phosphate synthase.

Claim 19 (Previously presented): The method of claim 1, wherein the target gene encodes a protein and the expression of said encoded protein is modulated.

Claim 20 (Currently amended): The method of claim 19, wherein the protein whose expression being modulated is heterologous to the plant cell tissue, ~~seed, or whole plant~~.

Claim 21 (Previously presented): The method of claim 20, wherein the protein whose expression being modulated is an antibody.

Claim 22 (Previously presented): The method of claim 19, wherein the expression of the protein is activated.

Claim 23 (Previously presented): The method of claim 19, wherein the protein whose expression being modulated participates in a metabolic pathway or controls a metabolic pathway.

Claim 24 (Previously presented): The method of claim 23, wherein the metabolic pathway is an anabolic or a catabolic pathway.

Claim 25 (Previously presented): The method of claim 23, wherein the metabolic pathway is for a molecule selected from the group consisting of a vitamin, a taste molecule, an anti-oxidant, a sugar and a flavanoid molecule.

Claim 26 (Previously presented): The method of claim 25, wherein the taste molecule is a bad taste molecule.

Claim 27 (Currently amended): The method of claim 23, wherein the metabolic pathway is heterologous to the plant cell ~~tissue, seed, or whole plant~~.

Claim 28 (Currently amended): The method of claim 23, wherein the metabolic pathway enhances an input or output trait in a plant cell ~~tissue, seed, or whole plant~~.

Claim 29 (Previously presented): The method of claim 19, wherein the target gene encodes an enzyme, a transport protein, a nutrient or storage protein, a contractile or motile protein, a structural protein, a defense protein or a regulatory protein.

Claim 30 (Previously presented): The method of claim 19, wherein the target gene encodes an enzyme or a co-factor in a metabolic pathway.

Claim 31 (Previously presented): The method of claim 1, which method is used for treating a disorder in the plant cells, wherein the disorder is associated with abnormal expression of the target gene.

Claims 32-35 (Canceled)

Claim 36 (Previously presented): The method of claim 1, wherein the zinc finger protein binds to the complementary strand of the target nucleotide sequence.

Claim 37 (Previously presented): The method of claim 1, wherein the zinc finger protein specifically binds to an effector domain of the target sequence and whereby the expression of the target gene is modulated by competitive inhibition of said effector domain.

Claim 38 (Previously presented): The method of claim 37, wherein the zinc finger protein does not comprise an effector domain.

Claim 39 (Currently amended): The method of claim 1, wherein the zinc finger protein comprises an effector domain active in the host plant cell ~~tissue, seed, or whole plant~~.

Claim 40 (Previously presented): The method of claim 1, wherein the zinc finger protein comprises a plurality of finger regions.

Claim 41 (Previously presented): The method of claim 40, which comprises linker regions among the plurality of finger regions.

Claim 42 (Currently amended): The method of claim 1, wherein the zinc finger protein comprises at least two 3-finger regions and the linker region between any said two 3-finger ~~region~~ regions is from about 2 to about 10 amino acid residues in length.

Claim 43 (Currently amended): The method of claim 42, wherein the linker region between any said two 3-finger ~~region~~ regions is about 5 amino acid residues in length.

Claim 44 (Previously presented): The method of claim 1, wherein the zinc finger protein comprises a framework from a plant zinc finger protein

Claim 45 (Canceled)

Claim 46 (Previously presented): The method of claim 1, wherein the zinc finger protein is selected from the group consisting of ZFPm1, ZFPm2, ZFPm3, ZFPm4 and ZFPAp3.

Claim 47 (Previously presented): The method of claim 1, wherein the zinc finger protein is not a zinc finger-nucleotide binding polypeptide variant comprising at least three zinc finger modules that bind to a target cellular nucleotide sequence and modulate the transcriptional function of the cellular nucleotide sequence, wherein the amino acid sequence of each zinc finger module that binds a target cellular nucleotide comprises two cysteines and two histidines

whereby both cysteines are amino proximal to both histidines and wherein each of three modules of said variant has at least one amino acid sequence modification.

Claim 48 (Currently amended): The method of claim 1, wherein the plant ~~tissue, seed, or whole plant~~ cell is a monocot or dicot plant ~~cell tissue, seed, or whole plant~~.

Claim 49 (Currently amended): The method of claim 1, wherein the plant cells ~~are~~ is included within an intact plant or constitutes all the cells of an intact plant.

Claim 50 (Previously presented): The method of claim 1, wherein the plant cells are protoplasts or spheroplasts.

Claim 51 (Previously presented): The method of claim 1, wherein the modulation of the gene expression is activation or repression.

Claim 52 (Previously presented): The method of claim 1, wherein the modulation of the gene expression is at least two fold.

Claim 53 (Previously presented): The method of claim 52, wherein the modulation is at least five fold repression.

Claim 54 (Previously presented): The method of claim 52, wherein the modulation is at least two fold activation.

Claim 55 (Currently amended): The method of claim 1, wherein the modulation changes the phenotype of the plant cell ~~tissue, seed, or whole plant~~.

Claim 56 (Previously presented): The method of claim 4, wherein the plant cells are contained in an *in vitro* culture.

Claim 57 (Currently amended): The method of claim 4, wherein the plant cell ~~is culturing cultured~~ is in planta.

Claim 58 (Previously presented): The method of claim 1, wherein the expression system comprises an inducible promoter.

Claim 59 (Previously presented): The method of claim 1, wherein the expression of the zinc finger protein is controlled by a tissue-specific promoter and whereby tissue-specific modulation of the target gene expression is obtained.

Claim 60 (Previously presented): The method of claim 59, wherein the tissue is selected from the group consisting of calli, meristem, leave, root and organ explant in tissue culture.

Claim 61 (Previously presented): The method of claim 1, wherein the zinc finger protein is expressed in a specific organelle.

Claim 62 (Previously presented): The method of claim 61, wherein the organelle is selected from the group consisting of a mitochondria, a nucleus, a plastid and a vacuole.

Claim 63 (Previously presented): The method of claim 62, wherein the plastid is selected from the group consisting of a chloroplast, a leucoplast, an aravloplast and a chromoplast.

Claim 64 (Currently amended): The method of claim 1, wherein the zinc finger protein is stably integrated in a specific organelle of a plant cell~~tissue, seed, or whole plant~~.

Claim 65 (Previously presented): The method of claim 1, wherein the zinc finger protein is targeted to a specific organelle.

Claim 66 (Previously presented): The method of claim 65, wherein the zinc finger protein is targeted to plastid via a plastid transit peptide, to chloroplast via a chloroplast transit peptide, to mitochondrial via a mitochondrial target peptide or to nucleus via a nuclear targeting peptide.

Claim 67 (Canceled)

Claim 68 (Currently amended): The method of claim 1, wherein the zinc finger protein comprises preferred codons of the host plant cell~~tissue, seed, or whole plant~~.

Claim 69 (Currently amended): The method of claim 68, wherein the zinc finger protein comprises preferred translational start codon of the host plant cell~~tissue, seed, or whole plant~~.

Claim 70 (Currently amended): A method of stably modulating a level of a compound in a plant cell ~~tissue, seed, or whole plant~~, which method comprises stably expressing in a plant cell ~~tissue, seed, or whole plant~~ a synthetic zinc finger protein that specifically binds to a target nucleotide sequence, wherein said target nucleotide sequence comprises 18 nucleotides and wherein said zinc finger protein is a hexadactyl zinc finger protein, wherein said zinc finger protein comprises one individual zinc finger DNA binding site with mutations at one or more of the base-contacting positions, within a target gene to modulate expression of said target gene which is involved in a compound's metabolism in said plant cell ~~tissue, seed, or whole plant~~, whereby the level of said compound in said plant cell ~~tissue, seed, or whole plant~~ is modulated.

Claim 71 (Previously presented): The method of claim 70, wherein the compound is phytic acid.

Claim 72 (Previously presented): The method of claim 70, wherein the target gene encodes AP3.

Claim 73 (Previously presented): The method of claim 70, wherein the zinc finger protein is not a zinc finger-nucleotide binding polypeptide variant comprising at least three zinc finger modules that bind to a target cellular nucleotide sequence and modulate the transcriptional function of the cellular nucleotide sequence, wherein the amino acid sequence of each zinc finger module that binds a target cellular nucleotide comprises two cysteines and two histidines whereby both cysteines are amino proximal to both histidines and wherein each of three modules of said variant has at least one amino acid sequence modification.

Claim 74 (Currently amended): An expression vector for stably modulating gene expression in ~~[[a]] plant cell tissue, seed, or whole plant~~, which expression vector comprises a nucleotide sequence encoding a synthetic zinc finger protein, wherein said zinc finger protein is a hexadactyl zinc finger protein, wherein said zinc finger protein comprises one individual zinc finger DNA binding site and has mutations at one or more of the base-contacting positions; said zinc finger protein is capable of specifically binding to a target nucleotide sequence, or a complementary strand thereof, within a target gene whose expression is to be stably modulated by said zinc finger protein in said plant cell tissue, seed, or whole plant.

Claim 75 (Previously presented): The expression vector of claim 74, wherein the zinc finger protein is not a zinc finger-nucleotide binding polypeptide variant comprising at least three zinc finger modules that bind to a target cellular nucleotide sequence and modulate the transcriptional function of the cellular nucleotide sequence, wherein the amino acid sequence of each zinc finger module that binds a target cellular nucleotide comprises two cysteines and two histidines whereby both cysteines are amino proximal to both histidines and wherein each of three modules of said variant has at least one amino acid sequence modification.

Claim 76 (Currently amended): A genetically modified plant ~~cell tissue, seed, or whole plant~~, which wherein plant cell tissue, seed, or whole plant comprises an expression system for a stably expressed synthetic zinc finger protein under the control of a first promoter, wherein said zinc finger protein is a hexadactyl zinc finger protein, wherein said zinc finger protein comprises one individual zinc finger DNA binding site with mutations at one or more of the base-contacting positions, said zinc finger protein is capable of specifically binding to a target nucleotide sequence, or a complementary strand thereof, within a target gene whose expression is to be stably modulated by said zinc finger protein.

Claim 77 (Currently amended): The genetically modified plant ~~cell tissue, seed, or whole plant~~ of claim 76, wherein the target nucleotide sequence is endogenous or exogenous to the targeted gene.

Claim 78 (Currently amended): The genetically modified plant cell ~~tissue, seed, or whole plant~~ of claim 76, wherein the target gene is endogenous or exogenous to the plant cell ~~tissue, seed, or whole plant~~.

Claim 79 (Previously presented): The genetically modified plant cell of claim 76, which is contained in an intact plant.

Claim 80 (Currently amended): The genetically modified plant cell ~~tissue, seed, or whole plant~~ of claim 76, wherein the zinc finger protein controls its own expression by binding to a target sequence within the zinc finger protein gene.

Claim 81 (Currently amended): The genetically modified plant cell ~~tissue, seed, or whole plant~~ of claim 76, wherein the zinc finger protein controls its own expression by binding to a first target sequence within the zinc finger protein gene and controls the expression of the target gene by binding to a second target sequence within the target gene.

Claim 82 (Currently amended): The genetically modified plant cell ~~tissue, seed, or whole plant~~ of claim 81, wherein the first target sequence within the zinc finger protein gene is different from the second target sequence within the target gene.

Claim 83 (Currently amended): The genetically modified cell ~~plant tissue, seed, or whole plant~~ of claim 76, wherein the zinc finger protein gene is further controlled by a second promoter.

Claim 84 (Currently amended): The genetically modified plant cell ~~tissue, seed, or whole plant~~ of claim 83, wherein the second promoter is inducible.

Claim 85 (Currently amended): The genetically modified plant cell ~~tissue, seed, or whole plant~~ of claim 76, wherein the zinc finger protein comprises at least ~~two~~ six zinc finger sequences.

Claims 86-87 (Canceled)

Claim 88 (Currently amended): The genetically modified plant cell tissue, seed, or whole plant of claim 76, which is selected from the group consisting of a tobacco, tomato, potato, banana, soybean, pepper, wheat, rye, rice, spinach, carrot, maize and corn

Claim 89 (Currently amended): The genetically modified plant cell tissue, seed, or whole plant of claim 76, wherein the zinc finger protein expressed in the plant cell tissue, seed, or whole plant is not a zinc finger-nucleotide binding polypeptide variant comprising at least three zinc finger modules that bind to a target cellular nucleotide sequence and modulate the transcriptional function of the cellular nucleotide sequence, wherein the amino acid sequence of each zinc finger module that binds a target cellular nucleotide comprises two cysteines and two histidines whereby both cysteines are amino proximal to both histidines and wherein each of three modules of said variant has at least one amino acid sequence modification.

Claim 90 (canceled)

Claim 91 (Currently amended): A genetically modified plant cell tissue, seed, or whole plant comprising an exogenous, synthetic zinc finger protein that specifically binds to a target nucleotide sequence in said plant cell tissue, seed, or whole plant wherein said exogenous zinc finger protein is constitutively expressed, wherein the target nucleotide sequence comprises 18 nucleotides and wherein said zinc finger protein is a hexadactyl zinc finger protein, wherein said zinc finger protein comprises one individual zinc finger DNA binding site with mutations at one or more of the base-contacting positions.

Claim 92 (Currently amended): A genetically modified plant cell tissue, seed, or whole plant comprising an exogenous, synthetic zinc finger protein that specifically binds to a target nucleotide sequence in said plant cell tissue, seed, or whole plant wherein said exogenous zinc finger protein is inducibly expressed, wherein the target nucleotide sequence comprises 18 nucleotides and wherein the zinc finger protein is a hexadactyl zinc finger protein, wherein said zinc finger protein comprises one individual zinc finger DNA binding site with mutations at one or more of the base-contacting positions.

Claim 93 (Previously presented): A genetically modified plant tissue, which tissue comprises the genetically modified plant cell of claim 76.

Claim 94 (Previously presented): A genetically modified plant seed, which seed comprises the genetically modified plant cell of claim 76.

Claim 95 (Previously presented): The genetically modified plant seed of claim 9476, which is selected from the group consisting of a tobacco, tomato, potato, banana, soybean, pepper, wheat, rye, rice, spinach, carrot, maize and corn seed.

Claim 96 (Canceled)

Claim 97 (Previously presented): A genetically modified plant seed, which seed comprises the genetically modified plant cell of claim 89.

Claim 98 (Currently amended): A plant that is regenerated from a plant cell transformed with the expression vector of claim 74.

Claim 99 (Currently amended): A method to stably modulate expression in a plant cell, which method comprises culturing the plant cell of claim 76.

Claim 100 (Previously presented): The method of claim 99, wherein the plant cell is cultured in planta.

Claim 101-132 (Canceled)

Claim 133 (Previously presented): The method of claim 1, wherein the zinc finger protein comprises a framework (or backbone) derived from a naturally occurring zinc finger protein.

Claim 134 (Previously presented): The method of claim 1, wherein the zinc finger protein comprises a framework (or backbone) derived from a zinc finger protein comprising a C2H2 motif.

Claim 135 (Previously presented): The method of claim 134, wherein the protein or peptide sequence within the β sheet of the C2H2 motif is not changed from its natural sequence.

Claim 136 (Previously presented): The method of claim 1, wherein the zinc finger protein comprises a framework (or backbone) derived from a zinc finger protein that is naturally functional in plant cells.

Claim 137 (Previously presented): The method of claim 136, wherein the framework (or backbone) comprises a motif selected from the group consisting of a C3H zinc finger, a QALGGH motif, a RING-H2 zinc finger motif, a 9 amino acid C2H2 motif, a zinc finger motif of Arabidopsis LSD1 and a zinc finger motif of BBF/Dof domain proteins.

Claim 138 (Previously presented): The method of claim 1, wherein the zinc finger protein comprises a framework (or backbone) derived from a zinc finger protein that is known in the art as of January 19, 2001.